REMARKS

Entry of the amendment is respectfully requested to reduce the number of issues on appeal.

Claims 27-29, 31, 33 and 41-54 are before the Examiner. Claims 1-26 and claims 34-36, directed to a nonelected invention, have been withdrawn from consideration by the Examiner pursuant to Rule 142(b). Claim 30 has been canceled and replaced with claim 54.

It is noted that claims 37-49, newly submitted in the previous response, were renumbered by the Examiner as claims 41-53.

Additionally, Examiner's concerns under 37 CFR 1.75(c) relating to claim 33 have been addressed by the amendments above.

The Section 112 Rejections

Claims 27-31, 33 and 41-54 are rejected under 35 USC 112, first paragraph, as containing subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventors, at the time the application was filed, had possession of the claimed invention. Applicants respectfully traverse.

Claim 27 is directed to a porous free-flowing fungal granulate, formed by a mechanical extrusion process, principally composed of intact dead microbial cells, where the granulate structure allows sufficient solvent access, via the pores, to the intact dead cells to isolate or extract a compound from the microbial cells.

Claim 28 is directed to a composition of granular particles having a dry matter content of at least 30% but less than 70% and a structure that on drying allows isolation or extraction of a compound from the intact dead cells with a solvent through pores and/or channels where the granulates comprise intact dead microbial cells obtained by granulating the biomass having a dry matter content of 25 to 80%.

Claim 54 (claim 30) is directed to a dried granular composition of dead microbial cells obtained by drying granular particles resulting from the granulation of a microbial biomass having a dry matter content of at least 80% and a structure that allows, via the pores, sufficient

solvent access to a compound contained within the intact dead cells to isolate or extract the compound therefrom.

These compositions of matter are clearly described in the specification. Population(s) of dead (killed) intact cells which have been processed to enhance solvent extraction/isolation of desired compounds.

The Examiner appears to be of the opinion that one skilled in the art would not consider an extrudate of intact dead cells to have been described in the application as originally filed. The basis for this appears to be the Examiner's opinion that extrusion must result under all conditions in cell rupture and therefore, a population of intact dead cells could not be produced. No reasoning for this is provided. This is clearly not the case.

It is clear from the enclosed Declaration by Hendrick Louis Bijl, an expert and also one of the coinventors, in paragraphs numbered 2 through 5, that extrusion does not have to result in the rupture of cells and that the avoidance of cell rupture under extrusion conditions is not difficult to achieve. Hendrick Louis Bijl indicates that conditions for achieving this end are taught within the specification. Further, Hendrick Louis Bijl clearly indicates that there is no inconsistency in the use of extrusion and obtaining a resultant intact cell product.

It is respectfully submitted that the claimed invention is clearly described to one skilled in the relevant art.

The use of granules and granulates are seen through out the specification as these terms are used frequently. Granulating (see page 1, line 12, page 4, line 13 *inter alia*) clearly would produce a granulate.

As for "intact", the specification makes it clear that damage to the cells should be minimized (page 5, lines 7 to 9 and page 6, lines 10-17) and the word "intact" itself appears on page 16 at line 15 and also on page 21 at line 7.

As for the Pichia organisms, the text the Examiner refers to merely states that for these organisms " preferably no pasteurization is conducted". This does not mean that pasteurization can never be preformed on Pichia organisms. Nor does it mean that Pichia cells can not be killed by some other means, or at some other stage during the process. For example, extrusion may take place at from 10 to 60° C (page 14, line 25) and at the top end of this range such high temperatures may be sufficient to kill Pichia cells. Similarly, in order to achieve the dry matter content of the claims, drying may take place, and with those temperatures such as those

described on page 18 at lines 28 and 29 again this may be sufficient to kill these cells. Therefore, cells can be killed by other methods in other processing steps before the compound is isolated or extracted therefrom.

Accordingly, withdrawal of the rejection is respectfully requested in light of the declaration the arguments above and a failure to establish a proper prima facie case in regards to the grounds for the rejection.

Claims 27-31, 33 and 41-54 are rejected under 35 USC 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. Applicants respectfully traverse.

Claim 30 has been rewritten as claim 54 to address the point(s) raised by the Examiner. Claim 33 has been amended to avoid the multiplicity issue raised by its dependency on claim 31.

Applicants have difficulty appreciating the Examiner's position relative to 1) the clarity of the phrase "granulate formed by extrusion which consists essentially of dead intact microorganisms or dead microbial cells", 2) the inconsistency between the use of extrusion and the description of resultant product as having "intact" cells and 3) solvent access "via the pores". It is not clear how these phrases obscure the boundaries of the claims as understood by one skilled in the relevant art.

There is no inconsistency between the use of extrusion and obtaining a resultant intact cell product. See declaration and comments, supra.

With regard to "solvent" access via pores, it is not clear how, based on the Examiner's stated position, a recitation of a mechanism of solvent entry (contact) would render the claims indefinite. The claim should be read in its entirety and in light of the specification. It is clear that the granulate is contacted with the appropriate solvent to isolate, recover, extract, etc. the desired compound from dead intact microbial cells. The invention is the "granulate" as claimed and the recognition that it results in the improved recovery of the desired product. It is not clear why the solvent would not enter through an orifice or why the specification of "pore" would render the boundary of the claims unclear in this regard.

The exemplary processes associated with "granulate formed by extrusion which consists essentially of dead intact microorganisms or dead microbial cells" are clearly identified. It is not

clear why this product defined in terms of its process of preparation would be considered indefinite in light of the claim format used. This claim form has long been excepted.

In light of the amendments, declaration and arguments, withdrawal of the rejection is respectfully requested.

The Art Rejections

Claims 27, 29, 30-32, and 37 are rejected under 35 USC 102 (e), as anticipated by Rhodes *et al.* (U.S. Patent No. 5,759,562). Applicants respectfully traverse.

The questioning of patentability in view of an alleged inconsistency regarding whether the cells are killed or intact is not appreciated. It is difficult to see why this affects patentability over Rhodes et al. The Applicant has already stated that the cells are dead in the independent claims, because they have been killed. This seems to have little bearing on whether the cells are intact or not, although in the present invention the cells do remain intact, since disruption of the cell wall is minimized. Simply because the cells are intact does not mean that they must be alive. Clearly, if the cells are not intact, that is to say that there has been massive disruption of the cell wall, then the cell will not survive and it will die. At a basic level therefore, disrupted or broken cells will be dead. However, that does not preclude the reverse situation, namely that dead cells can be intact. There are various ways to kill a cell, but that does not necessarily result in disruption of the cell wall and therefore death.

The claims, due in part to the "consisting essentially of " language are directed to a population of dead intact cells, processed in a manner so that there is enhance recovery of a desired product by solvent extraction or solvent isolation. Live cells would materially change the characteristic of the invention. There is evidence for this already in the specification. For example, one of the reasons that the cells are killed is that this inactivates one or more enzymes that could adversely affect the yield of that compound. For example, pasteurization by heating may inactivate enzymes that degrade the very desired compound that one is intending to extract. Therefore, in the context of the present invention, there is an appreciable and significant difference between the cells being alive and dead.

The Examiner suggests that there is nothing on record to suggest that to a person of skill in the art the prior art materials are not ones from which one can readily extract commercial amounts of intracellular compounds. With respect, this is not true. This can be readily seen

from several of the prior art documents.[1] As for Rhodes et al., there is absolutely no motivation to extract an entomopathogenic compound from the granular composition. Rhodes teaches that the fungi must be alive in order that they grow and sporolate in the soil. In order to extract a compound from the fungus, this might involve killing the organism, which would defeat the main purpose of providing this fungus according the Rhodes. For this reason alone there would be a considerable disincentive to extract the active compound. Indeed, Rhodes et al do not identify any particular compound that is active against pests such as insects. There is not mention anywhere of there being a particular agent, produced by the fungi, that is pathogenic to soil pests. It is therefore entirely speculative for the Examiner to suggest that one single compound, for example one that is entomopathogenic, actually exists. Indeed, the reason why compositions of Rhodes et al. appear to be effective is that they grow and sporolate in the soil, and are present in the form of blastospores. There is no suggestion anywhere that entomopathogenic compounds exist, or are produced by fungi, let alone that it would be useful to extract such a compound. There is therefore no evidence in Rhodes et al to support the Examiner's speculation. Furthermore, although the Examiner suggest that there may be compelling motivation to solvent extract such a compound, there is no evidence of any such motivation in Rhodes et al. Even if such compounds did exist, there is no suggestion for their speculation on what a skilled person might be motivated to do is lacking in scientific basis in evidence, and is nothing more than speculation and hindsight.

Rhodes *et al.* clearly teaches a composition containing an insecticidal amount of blastophores and a complex nutrient source for application to soil to control soil dwelling insect pests. A blastophore is a spore produced by a budding process along the mycelium or by a single spore. A spore is a resistant body formed by certain microorganisms, a resistant resting cell; a primitive unicellular reproductive body. Spores are not the materials from which one readily extracts "commercial" amounts of intracellular compounds, e.g. β-carotene, Vitamin B-12, etc.

A spore and nutrient composition teaching is simply not a teaching of a granulated intact fungal cells having a porous structure which facilitates the extraction of intracellular compounds. The microbial cells are dead (killed).

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[1] For example, Huang et al. describes fungal masses that have been thermal shocked to reduce the nucleic acid content before being formed into fiber bundles for eating (as a substitute for meat). Cockram et al. similarly refers to the formation of meat-like materials.

Additionally, claim 27 specifies that the granules are porous and/or have hollow channels. These pores and channels provide access into the center of the granules (see page 20, starting at line 11 and continuing to the first five lines of page 21). Further, the granules are free flowing (see page 19, line 28 and page 20 at line 4). As already noted, "consisting essentially of" further excludes the Rhodes *et al.* compositions that are directed to live cells. Please also note that the dried granules have a porous structure that allows access for the solvent to the dead cells to extract a desired compound therefrom. Basis for this can be found in the paragraphs spanning pages 6 and 7, on page 14 (first four lines) and on page 20 at line 31.

Withdrawal of the rejection is respectfully requested.

Claims 28-33 and 30-40 are rejected under 35 USC 102(b) as anticipated by or, in the alternative, under section 103(a), as obvious over Huang *et al.* or Cockram *et al.* Applicants respectfully traverse.

The three independent claims, as amended, specify that the structure of the granulate and dry granules allow, via the pores and/or channels, access of a solvent to the dead cells so that one can isolate or extract the "desired" compound therefrom. The same is true of the granular particles of claim 29, although they require drying first (to give the dry granules).

Further, the present application teaches numerous advantages of granulation techniques in order to efficiently permit extraction of desired compounds from biomass or fungal cell materials. The benefits are illustrated in the Examples, Tables and Figures. In particular, consider Tables 2 and 5; Example 25 in contrast to Comparative Example 26; and Figures 5 and 6; Table 5, Example 25 in contrast to Comparative Example 26. These comparisons illustrate the benefits of the granulated/extruded biomass. Table 2 illustrates the effects of different dry matter contents on the quality of the extrudate. Figures 5 and 6 illustrate the enhanced porosity of the claimed product.

According to the practice of the present invention, granular particles are formed from biomass <u>having a dry matter content from 25 to 80%</u>. Following drying of the particles, desired compounds are extracted therefrom. Granular particle size, water content of the biomass and its control at various stages of the process are critical to achieving the claimed granular structures which possess numerous advantages. The resultant dried granules permit maximum solvent access for extraction while at the same time avoiding fines or dust from milling that may impede filtration. Granules permit more efficient extractions than larger particles such as flakes. (See

the Specification at page 4, lines 6-32; page 7, line 6; page 13, lines 29-32; page 20, lines 33-36.) Further, damage to fungal cells is minimized when granule formulations are prepared, such as by extrusion, and there is generally no need to disrupt cells prior to extraction of desired compounds. (See page 5, lines 7-9; page 6, lines 10-17; page 14, lines 8-11; page 21, line 7; page 7, line 10.) This permits the granular biomass to be storage-stable which allows "breaks" in processing, e.g., drying of the extruded granules does not have to be immediately undertaken after their preparation.

None of these advantages (or other advantages referred to in the Specification) are at all evident from Huang *et al.* or Cockram *et al.*, alone or in combination.

The Examiner suggests that there is nothing on record to suggest that to a person of skill in the art the prior art materials are not ones from which one can readily extract commercial amounts of intracellular compounds. With respect, this is not true. This can be readily seen from several of the prior art documents. For example, Huang et al. describes fungal masses that have been thermal shocked to reduce the nucleic acid content before being formed into fiber bundles for eating (as a substitute for meat). Cockram et al. similarly refers to the formation of meat-like materials.

Cockram *et al.* deals with the texturizing of a mycelium fungal mass containing 20% to 35% solids in a water base. There is no drying step mentioned. A solvent-extracted compound is not mentioned either. The process described does not produce granules. Rather, it forms strands from filaments containing mycelium mass. The strands are incorporated into a food stuff. There is clearly no disclosure of the need to enhance porosity of fungal cells or how to do it.

Dealing first with the Cockram *et al.* document, this refers to texturizing a mycelial fungal mass by extruding fungi through a die under high pressure (column 2, lines 24-25). This forms axially parallel filaments (column 2, lines 66-67). These are not granules. In addition, there is no disclosure that the fungal cells must be dead as required by the claims. The process of this document could easily be conducted on live cells. There is nothing in the Cockram *et al.* document that teaches that the composition is either free flowing or is granular.

Huang is directed to the treatment of a proteinaceous mass of fungal fibers by rapid dielectric heating in order to reduce the amount of nucleic acid therein. The end product is one that has the texture and chewability of meat. This is not akin to the extraction of Vitamin B-12.

The Applicants note the Examiner's comments about the U.S. Patent and Trademark Office not being equipped to manufacture products by prior art methods and then to compare those with the products now claimed. However, before Applicants should be required to undertake the expense of a side-by-side comparison, the products should reasonably appear to be the same. That is not the case here. The different purpose(s) of the prior art suggest preparation and product differences from that claimed herein. The presence of inherent characteristics must be certain and not based on mere speculation.

Neither document specifically discloses granules that have a structure that allows a solvent access, via the pores, to the dead cells in order to isolate or extract desired compounds therefrom. Both of these documents deal with texturizing fungi, in particular to produce filamentous products or fiber bundles. There is no intention to extract a compound from these elongate extrudates. There is therefore no reason to suppose that they have a structure that allows solvent access. Indeed, neither of them discloses that the products are porous, nor that the structure of the pores allows solvent access to the dead cells.

The Examiner has not specified where in either of these two documents all of the features of the claims can be found. Instead, the Examiner is merely speculating that as both documents refer to extrusion of a fungal biomass then one must obtain the same product as now presently claimed. That is not true, and it is a false assumption to make. The Examiner has no evidence that the prior art products would have the same feature as the granules of the present application, especially as they have different uses and purposes, and therefore neither document can be considered to be novelty destroying.

Further, the specification in the passages referred to above illustrate benefits for the claimed invention that are not expected from the cited references.

It is also submitted that both of the cited documents are entirely in different fields, and while they all refer to fungal biomass, there the commonality ends: the uses of the biomass are very different.

The problem that a skilled artisan faces in the present application is to efficiently extract a desired compound from a biomass. The disclosed invention achieves this by extruding the (already killed) intact cells into a granular form which is porous and has hollow channels. These pores and channels allow access of the solvent to the dead intact fungal cells and because a large

surface area is provided to the solvent, the desired compound can be extracted from those cells with great efficiency. None of the prior art documents extrude, or form granulates.

It is not seen why the Cockram and Huang processes, which are clearly in different fields of endeavor and have different ends in mind than that of the invention, would reasonably be expected to result in a product having the claimed characteristics.

For a start, both of these documents intend to produce textured fungal products for human consumption. These substances are incorporated into a foodstuff. In contrast, the granules of the present application are not intended to be edible; instead, they are to be treated with a solvent in order to isolate a compound therefrom. That places a very different emphasis on the different products and their uses.

The art produces parallel filaments or mycelial fibers; in the case of Cockram *et al.*, the products remain fibrous (they are not granular). This is to provide fibrous texture. In contrast, the granules of the present application (which are not fiber bundles) possess pores to allow access of solvent for compound extraction. Indeed, even after the parallel filaments are formed, further extrusion forms the filaments into strands (column 3, lines 10-11) so at all times there are continuous threads or filaments rather than granules.

While the Examiner may have concerns that the products of the prior art might be similar to those claimed, it is clear from the very different uses that it is likely, beyond reasonable doubt, that the granules of the present application have a different structure from those described in the prior art.

Since neither a proper *prima facie* case of anticipation or obviousness has been established in regard to the claims as amended; and further, since the results achieved by the claimed invention would not be expected from the applied prior art, withdrawal of the rejection is respectfully requested.

Claims 42-49 are rejected under 35 USC 103(a) as being unpatentable over Huang *et al.* or Cockram *et al.* or Rhodes *et al.* and further in view of Akimoto et al.. Claims 50-51 are rejected under 35 USC 103(a) as being unpatentable over Huang *et al.* or Cockram *et al.* or Rhodes *et al.* and further taken with Casey *et al.* Claims 52-53 are rejected under 35 USC 103(a) as being unpatentable over Huang *et al.* or Cockram *et al.* or Rhodes *et al.* and further taken with Rickes. Applicants respectfully traverse.

The deficiencies of the principle references are not remedied by the additional secondary references, taken alone or in combination.

Akimoto et al or Casey et al. provide nothing more than suggesting that a PUFA can be extracted from a fungus (Akimoto) or that TAPS can be extracted from a strain of Pichia. These two documents would not be combined with any of Rhodes, Cockram or Huang because they are entirely different fields. Huang and Cockram both deal with edible meat substitutes. Rhodes deals with insecticidal compositions containing fungi. This has nothing in common with extracting compounds from microorganisms as described by Akimoto or Casey. The two fields of art are entirely different, as illustrated by the International and US classifications allotted to these two groups of documents. A person skilled in the art looking to improve on the extraction of a particular compound from a microorganism would not look to documents that concern either meat substitutes or insecticides. It is therefore certainly not obvious to modify the teachings of one group of documents with the other group. Indeed, the fact that the present Applicant has discovered that efficient extraction can be achieved with the compositions now claimed illustrates that he has taken some, but not all, aspects of technology from very different fields of endeavor. It is certainly not obvious to combine meat substitutes or insecticides with Akimoto or Casey et al. Indeed, to make such a combination one would require hindsight. The Examiner is only combining these two fields of art now that he has seen the Applicants current invention. The Examiner can therefore only make the combination he has done in the Office Action with the benefit of hindsight, which it is suggested is inadmissible in the present case.

Likewise, Rickes, which teachings, if taken as the states, merely describes bacteria as a source of Vitamin B-12 that can be solvent extracted. This teaching does not remedy the deficiencies noted above with regard to the preparation of the granulate and/or its characteristics.

Withdrawal of the rejections is respectfully requested.

CONCLUSION

Having addressed all the objections and rejections, the application is believed to be in condition for allowance. A notice to that effect is respectfully requested.

In the unlikely event that the transmittal letter is separated from this document and the Patent Office determines that an extension and/or other relief is required, Applicants petition for

any required relief including extensions of time and authorize the Assistant Commissioner to charge the cost of such petitions and/or other fees due in connection with the filing of this document to **Deposit Account No. 03-1952** referencing docket no. 251502006900. However, the Assistant Commissioner is not authorized to charge the cost of the issue fee to the Deposit Account.

Respectfully submitted,

Dated: January 8, 2001 By: _____

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IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In the application of:
Hendrik L. BIJL *et al*
Serial No.: 08/821,025
Filing Date: March 19, 1997
For: PROCESS FOR THE PREPARATION OF A GRANULAR MICROBIAL BIOMASS AND ISOLATION OF A COMPOUND THEREFROM

Examiner: Dr. Irene Marx
Group Art Unit: 1651

DECLARATION

I, Hendrik Louis BIJL, of Insulindestraat 72, Vlaardingen, The Netherlands, do hereby declare as follows:

1. I am the first named inventor on the above US patent application. I have read the Office Action of 6 July 2000, and the prior art documents referred to by the Examiner. I have been asked to comment on the matters raised by the Examiner.

2. As an inventor of the present application, I believe that I can clarify what would be understood by a person skilled in this art. The invention relates to the extraction of desired compounds from microbial biomass. The extraction takes place after the biomass has been processed into granules, for example by extrusion. Thus, this invention is in the field of isolating valuable compounds from microorganisms: this seems to me to be an entirely different field from that concerning textured biomass and meat substitutes and live fungal compositions to kill soil pests such as insects.

3. The Examiner appears to suggest that there is an inherent inconsistency between the preparation of the granulate (by killing the cells and submitting them to extrusion) and describing the cells as "intact". In my view these two elements are not inconsistent, indeed they may co-exist. In fact, such co-existence occurs in the invention that is the subject of the present application. That is to say, it is possible to granulate the biomass (after killing the cells, and for example using an extruder) and yet allow the cells to remain intact. This is achievable by using granulation techniques and conditions that do not disrupt the cell walls. This is described in the subject application, and it is not particularly difficult to keep the cells intact following the granulation process.

4. Perhaps the Examiner believes that when one kills the cells, this must inevitably result in disruption of the cell wall. This is not so. One can kill the cells by a variety of means, such as by heat, but that will not necessarily result in the cell walls becoming broken, and the cell splitting open. It is quite feasible to kill the cell, and yet the cell remain intact.

5. Similarly, it is feasible to granulate the biomass, again without breaking the cells. Extrusion is just one example of a granulation technique that will form granules but will leave the cells intact, that is to say without breaking the cell walls. That is precisely what happens in the present application, because in my invention it is advantageous for the cells to stay intact. If the cell walls were broken, then this would release the contents of the cell. That would make it much more difficult to isolate the desired compound, since then there would be a myriad of compounds from which the desired one would need to be extracted from. Keeping the cell walls intact can minimise, if not avoid, this problem altogether. There is therefore a not insignificant advantage in keeping the cells intact. Furthermore, the cells can be kept intact despite the fact that they have been killed and then granulated.

6. Turning to the prior art, both the Cockram *et al* and Huang *et al* documents seem very distant to my invention, and in a different technical field. Both relate to texturising fungi in order to prepare a meat-like material. Neither document aims to isolate compound from a microorganism, and therefore the texturising and other process steps are not designed to produce granulates of the type in my invention. Indeed, neither document appears to disclose the extraction of any compound from the fungal cells, they are merely concerned with providing meat substitutes. I would not expect either of these documents to disclosure granulates of the type now claimed, for example there is no evidence from either of these documents that the granulates have pores which would allow access of the compound to the cells inside the granulate, to allow extraction of a compound therefrom. Given that both of these documents aim to provide texturised meat substitutes, which is an entirely different purpose, it is perhaps not surprising that the compositions disclosed in these two documents are different from the granulate now claimed.

7. As for Rhodes *et al*, this describes compositions that are active against soil pests such as insects. They contain fungi that can grow and sporulate in the soil. This means that the fungi must be alive. In contrast, in my invention the cells must be dead before extraction of the desired compound can take place. The Examiner has apparently suggested that in Rhodes *et al* some of the fungi could be dead. I find no evidence in this document to

support such a statement. Indeed, Rhodes *et al* teaches that the fungi must be alive, if they are to form blastospores. From my point of view, this document clearly advocates that the fungi must be alive. There would be no advantage in providing fungi that are dead. In my view therefore the statement that some of the fungi could be dead is merely speculative, and it does not appear to be supported by any statement in the Rhodes *et al* citation. *In addition , this patent shows that cells after an extrusion step are intact and viable.*

8. The Huang *et al* and Cockram *et al* documents are both concerned with texturising fungi to provide meat substitutes. This, in my view, is a totally different field from the extraction of compound from microorganisms. Similarly, Rhodes *et al* deals with insecticides, containing live fungi, and again this would seem to have nothing to do with isolating desired compounds from dead intact fungal cells. I can see no connection between any of these three documents and the concept of extracting a desired compound from a microbial biomass granulate. It is unlikely that I would have turned to any of these documents, or other documents in this field, when initially thinking about how one could improve the yield and extraction of a particular compound from a microorganism.

I hereby declare that all the statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true. I further acknowledge that these statements are made in the knowledge that wilful false statements may jeopardise the validity of the application or any patent issued thereon.

Date: 04 January 2001

Hendrik Louis BIJL